IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:	)	Group Art Unit: 1637
Lau et al.	)	Examiner: Bertagna, Angela M.
Serial No.: 10/780,963	)	
Filed: Feb. 18, 2004	)	
For: Polyelectrolyte-Coated Size-Exclusion Ion-Exchange Particles	)	
Confirmation No.: 1685	)	

Declaration of Aldrich N.K. Lau Under 37 CFR 1.131

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

I, Aldrich N. K. Lau, declare and affirm as follows:

1. I am a Scientific Fellow in Research and Development for the Applied Biosystems, LLC in Foster City, California.

2. I hold a Ph.D. in synthetic organic chemistry, held a three year appointment as a Postdoctoral Fellow, and have 27 years experience in R&D in the private sector. I am the author or coauthor of over 19 scientific publications, as well as a named inventor on 18 issued US patents, as well as 24 US published patent applications.

3. I am an inventor on the present patent application (ASN 10/780,963).

4. I am familiar with the field of materials science related to biological analysis and during my work at Applied Biosystems, and I have become highly familiar with the field of the preparation of polynucleic acid-containing samples for analysis.

5. I have been advised that in an Office Action dated September 12, 2008, that claims 21, 22, 24, 45, 46, 48, 49, 66-69, and 76-79 have been rejected for being anticipation by Hennessey et al. (US Patent Appl. Pub. 2004/0016702; hereafter Hennessey '702). I have reviewed the rejected claims, of which the full set of currently pending claims of the present application is available as Exhibit 1.

6. I am highly familiar with Hennessey '702, as I am a named inventor on that application. Given my expertise in polymer science, I am the inventor of the polyelectrolyte coated particles of the present application, as well as the encapsulated particles of Hennessey '702. It should be noted that the two types of particles are prepared differently, and have distinctly different properties.

7. In the provisional patent application of Hennessey (US 60/398,852; filed July 26, 2002; hereafter Hennessey '852), I taught embodiments of an encapsulated particle having a neutral shell (see Exhibit 2; Fig. 1 from Hennessey '852). In the Action, reference is made by the Examiner regarding paragraph 68 of Hennessey '702 teaching embodiments of particles having a shell of a copolymer containing charged monomers. Given that this teaching was absent in Hennessey '852, it was then added at the time of filing of Hennessey '702, which has a filing date of August 11, 2003.

8. My work as an inventor of various embodiments of the polyelectrolyte particles of the present application and the encapsulated particles that I taught in Hennessey '702 was done in overlapping periods of time. For example, a notebook entry for the preparation of a copolymer for use as a coating of various embodiments of the polyelectrolyte particles of the present application is shown in Exhibit 3, which is work that predated the filing of Hennessey '702. The teachings of Exhibit 3 can be found, for example, but not limited by, in paragraph 73 of the present application. Later work on various embodiments of polyelectrolyte particles of the present application is shown in Exhibit 4, which also predated the filing date of Hennessey '702. The teachings of Exhibit 4 can be found, for example, but not limited by, in paragraphs 74 and 75 of the present application. Finally, my continued work on various embodiments of polyelectrolyte particles resulted in the filing of the present application on February 18, 2004. Accordingly, my work on reducing to practice various embodiments of the polyelectrolyte particles of the present application; especially as claimed in independent claims 21 and 45, was done before the filing of Hennessey '702.

9. I declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further, that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

January 14, 2009
Date

Aldrich N. K. Lau

Listing of Claims:

This listing of claims will replace all prior versions, and listings, of claims in the application:

What is claimed:

1. (Withdrawn) A particle comprising:

   a core comprising ion-exchange material; and

   a coating comprising polyelectrolyte material,

   wherein the core and coating are adapted to separate PCR reaction products.

2. (Withdrawn) The particle of claim 1, wherein the core couples to at least one PCR reaction product chosen from primers, primer-dimer, ssDNA fragments, unincorporated nucleotides, and salts.

3. (Withdrawn) The particle of claim 2, wherein the particle is adapted to substantially exclude dsDNA fragments having greater than 100 basepairs.

4. (Withdrawn) The particle of claim 1, wherein the coating comprises a biopolymer.

5. (Withdrawn) The particle of claim 4, wherein the biopolymer is non-sample DNA.

6. (Withdrawn) The particle of claim 1, wherein the coating comprises a synthetic polymer.

7. (Withdrawn) The particle of claim 6, wherein the synthetic polymer comprises a copolymer, wherein the copolymer comprises at least one monomer chosen from (meth)acrylamide, N-methyl (methyl)acrylamide, N,N-dimethyl (methyl)acrylamide, N-ethyl (meth)acrylamide, N-n-propyl (meth)acrylamide, N-iso-propyl (meth)acrylamide, N-ethyl-N-methyl (meth)acrylamide, N,N-diethyl (meth)acrylamide, N-hydroxymethyl (meth)acrylamide, N-(3-hydroxypropyl) (methy)acrylamide, N-vinylformamide, N-vinylacetamide, N-methyl-N-vinylacetamide, vinyl acetate (precursor of vinyl alcohol), 2-hydroxyethyl (meth)acrylate, 3-hydroxypropyl (meth)acrylate, N-vinypyrrolidone, poly(ethylene oxide) (methy)acrylate, N-(meth)acryloxysuccinimide,

   N-(meth)acryloylmorpholine, N-2,2,2-trifluoroethyl (meth)acrylamide,

N-acetyl (meth)acrylamide, N-amido(meth)acrylamide, N-acetamido (meth)acrylamide, N-tris(hydroxymethyl)methyl (meth)acrylamide, styrenesulfonic acid, homopolymers of styrenesulfonic acid, co-polymers of styrenesulfonic acid, N-(methyl)acryloyltris(hydroxymethyl)methylamide, (methyl) acryloylurea, vinyloxazolidone, vinylmethyloxazolidone, acrylic acid, methacrylic acid, vinyl sulfonic acid, styrene sulfonic acid, 4-acetoxystyrene (precursor of 4-hydroxystyrene), and vinylphosphonic acid, and vinyl methyl ether.

8. (Withdrawn) The particle of claim 7, wherein the synthetic polymer is poly(acrylic acid-co-N,N-dimethylacrylamide) or poly(N,N-dimethyl acrylamide-co-styrene sulfonic acid).

9. (Withdrawn) The particle of claim 1, wherein the ion-exchange material is porous.

10. (Withdrawn) The particle of claim 9, wherein the ion-exchange material is surface-activated.

11. (Withdrawn) The particle of claim 9, wherein the ion-exchange material has a pore size of 100 Angstroms to 2000 Angstroms.

12. (Withdrawn) The particle of claim 11, wherein the polyelectrolyte material has a Mw of 1.0 megaDaltons to 3.0 megaDaltons.

13. (Withdrawn) The particle of claim 12, wherein the ion-exchange material has the pore size of 1000 Angstroms and the Mw of 1.7 megaDaltons to 2.4 megaDaltons.

14. (Withdrawn) The particle of claim 6, wherein the synthetic polymer comprises a copolymer, wherein the copolymer comprises at least one monomer chosen from allyl amide hydrochloride, (3-acrylamidopropyl)trismethylammonium chloride, N-(3-aminopropyl)methacrylamide hydrochloride, and N-vinyl amides hydrolyzed to give an amino group.

15. (Withdrawn) The particle of claim 14, wherein the synthetic polymer is poly(N-(3-aminopropyl)methacrylamide-co-N,N-dimethylacrylamide).

16. (Withdrawn) The particle of claim 1, wherein the polyelectrolyte material comprises polyanions and polycations.

17. (Withdrawn) The particle of claim 16, wherein the polyanions and polycations form alternating layers.

18. (Withdrawn) A mixture comprising particles of claim 1, wherein the mixture includes a cationic ion-exchange material, and an anionic ion-exchange material.

19. (Withdrawn) A purification device comprising a receptacle, and the mixture of claim 18 disposed in the receptacle.

20. (Withdrawn) A microfluidic device comprising a plurality of columns, and the mixture of claim 18 disposed in each column.

21. (Previously Presented) A method for purifying PCR reaction products, the method comprising:

providing a plurality of particles, wherein each particle comprises an ion-exchange core coated by exposing the core to a polyelectrolyte copolymer material, wherein the polyelectrolyte copolymer material comprises at least one type of charged monomer and at least one type of neutral co-monomer;

providing a mixture of cationic ion-exchange particles and anionic ion-exchange particles, wherein the plurality of particles are either the cationic ion-exchange particles or the anionic ion-exchange particles; and

contacting the PCR reaction products with the mixture of particles to separate dsDNA fragments and purifying the PCR reaction products.

22. (Original) The method of claim 21, wherein the contacting comprises moving the PCR reaction products through the plurality of particles using centripetal force.

23. (Original) The method of claim 21, wherein the plurality of particles comprise a first volume, the PCR reaction products comprise a second volume, and the first volume is greater than or equal to the second volume.

24. (Original) The method of claim 21, further comprising positioning a mixture comprising the plurality of particles in a column.

25. (Withdrawn) A particle comprising:

a core comprising ion-exchange material; and

a coating comprising polyelectrolyte material,

wherein the core and coating are adapted to separate DNA sequencing reaction products.

26. (Withdrawn) The particle of claim 25, wherein the core couples to at least one DNA sequencing reaction product chosen from primers, dye-labeled primers, nucleotides, dye-labeled nucleotides, dideoxynucleotides, dye-labeled dideoxynucleotides, and salts.

27. (Withdrawn) The particle of claim 26, wherein the particle is adapted to substantially exclude dye-labeled ssDNA fragments having greater than 45 nucleotides.

28. (Withdrawn) The particle of claim 25, wherein the coating comprises a biopolymer.

29. (Withdrawn) The particle of claim 28, wherein the biopolymer is non-sample DNA.

30. (Withdrawn) The particle of claim 25, wherein the coating comprises a synthetic polymer.

31. (Withdrawn) The particle of claim 30, wherein the synthetic polymer comprises a copolymer, wherein the copolymer comprises at least one monomer chosen from (meth)acrylamide, N-methyl (methyl)acrylamide, N,N-dimethyl (methyl)acrylamide, N-ethyl (meth)acrylamide, N-n-propyl (meth)acrylamide, N-iso-propyl (meth)acrylamide, N-ethyl-N-methyl (meth)acrylamide, N,N-diethyl (meth)acrylamide, N-hydroxymethyl (meth)acrylamide, N-(3-hydroxypropyl) (methy)acrylamide, N-vinylformamide, N-vinylacetamide, N-methyl-N-vinylacetamide, vinyl acetate (precursor of vinyl alcohol), 2-hydroxyethyl (meth)acrylate, 3-hydroxypropyl (meth)acrylate, N-vinypyrrolidone, poly(ethylene oxide) (methy)acrylate, N-(meth)acryloxysuccinimide,

N-(meth)acryloylmorpholine, N-2,2,2-trifluoroethyl (meth)acrylamide,

N-acetyl (meth)acrylamide, N-amido(meth)acrylamide, N-acetamido (meth)acrylamide, N-tris(hydroxymethyl)methyl (meth)acrylamide, styrenesulfonic acid, homopolymers of styrenesulfonic acid, co-polymers of styrenesulfonic acid, N-(methyl)acryloyltris(hydroxymethyl)methylamide, (methyl) acryloylurea, vinyloxazolidone, vinylmethyloxazolidone, acrylic acid, methacrylic acid, vinyl sulfonic acid, styrene sulfonic acid, 4-acetoxystyrene (precursor of 4-hydroxystyrene), and vinylphosphonic acid, and vinyl methyl ether.

32. (Withdrawn) The particle of claim 31, wherein the synthetic polymer is poly(acrylic acid-co-N,N-dimethylacrylamide) or poly(N,N-dimethyl acrylamide-co-styrene sulfonic acid).

33. (Withdrawn) The particle of claim 30, wherein the synthetic polymer comprises a copolymer, wherein the copolymer comprises at least one monomer chosen from allyl amide hydrochloride, (3-acrylamidopropyl)trismethylammonium chloride, N-(3-aminopropyl)methacrylamide hydrochloride, and N-vinyl amides hydrolyzed to give an amino group.

34. (Withdrawn) The particle of claim 33, wherein the synthetic polymer is poly(N-(3-aminopropyl)methacrylamide-co-N,N-dimethylacrylamide).

35. (Withdrawn) The particle of claim 25, wherein the ion-exchange material is porous.

36. (Withdrawn) The particle of claim 35, wherein the ion-exchange material is surface-activated.

37. (Withdrawn) The particle of claim 35, wherein the ion-exchange material has a pore size of 5 Angstrom to 1000 Angstroms.

38. (Withdrawn) The particle of claim 37, wherein the polyelectrolyte material has a Mw of 1000 Daltons to 6.0 megaDaltons.

39. (Withdrawn) The particle of claim 38, wherein the ion-exchange material has the pore size of 10 Angstroms to 50 Angstroms and the Mw of 2.4 megaDaltons to 4.9 megaDaltons.

40. (Withdrawn) The particle of claim 25, wherein the polyelectrolyte material comprises polyanions and polycations.

41. (Withdrawn) The particle of claim 40, wherein the polyanions and polycations form alternating layers.

42. (Withdrawn) A mixture comprising particles of claim 25, wherein the mixture includes a cationic ion-exchange material, and an anionic ion-exchange material.

43. (Withdrawn) A purification device comprising a receptacle, and the mixture of claim 42 disposed in the receptacle.

44. (Withdrawn) A microfluidic device comprising a plurality of columns, and the mixture of claim 42 disposed in each column.

45. (Previously Presented) A method for purifying DNA sequencing reaction products, the method comprising:

providing a plurality of particles, wherein each particle comprises an ion-exchange core coated by exposing the core to a polyelectrolyte copolymer material, wherein the polyelectrolyte copolymer material comprises at least one type of charged monomer and at least one type of neutral co-monomer;

providing a mixture of cationic ion-exchange particles and anionic ion-exchange particles, wherein the plurality of particles are either the cationic ion-exchange particles or the anionic ion-exchange particles; and

contacting the DNA sequencing reaction products with the mixture of particles to separate dye-labeled ssDNA fragments and purifying the DNA sequencing reaction products.

46. (Original) The method of claim 45, wherein the contacting comprises moving the DNA sequencing reaction products through the plurality of particles using centripetal force.

47. (Previously Presented) The method of claim 45, wherein the plurality of particles comprise a first volume, the DNA sequencing reaction products comprise a second volume, and the first volume is less than or equal to the second volume.

48. (Original) The method of claim 45, further comprising removing residual dye artifacts.

49. (Original) The method of claim 45, further comprising maintaining dye-labeled ssDNA fragment length.

50. (Withdrawn) A method for forming a particle, the method comprising:

selecting core material and polyelectrolyte material adapted to separating at least one of PCR reaction products and DNA sequencing reaction products;

providing the core comprising ion-exchange material; and

coating the core with polyelectrolyte material.

51. (Withdrawn) The method of claim 50, further comprising activating the surface of the core.

52. (Withdrawn) The method of claim 50, further comprising rinsing excess polyelectrolyte material.

53. (Withdrawn) A composition comprising:

polyelectrolyte material wherein the polyelectrolyte material is adapted to coating ion-exchange material and to providing separation of at least one of PCR reaction products or DNA sequencing reaction products.

54. (Withdrawn) The composition of claim 53, wherein the polyelectrolyte material comprises a synthetic polymer.

55. (Withdrawn) The composition of claim 53, wherein the synthetic polymer comprises a copolymer, wherein the copolymer comprises at least one monomer chosen from (meth)acrylamide, N-methyl (methyl)acrylamide, N,N-dimethyl (methyl)acrylamide, N-ethyl (meth)acrylamide, N-n-propyl (meth)acrylamide, N-iso-propyl (meth)acrylamide, N-ethyl-N-methyl (meth)acrylamide, N,N-diethyl (meth)acrylamide, N-hydroxymethyl (meth)acrylamide, N-(3-hydroxypropyl) (methy)acrylamide, N-vinylformamide, N-vinylacetamide, N-methyl-N-vinylacetamide, vinyl acetate (precursor of vinyl alcohol), 2-hydroxyethyl (meth)acrylate, 3-hydroxypropyl (meth)acrylate, N-vinypyrrolidone, poly(ethylene oxide) (methy)acrylate, N-(meth)acryloxysuccinimide,

N-(meth)acryloylmorpholine, N-2,2,2-trifluoroethyl (meth)acrylamide,

N-acetyl (meth)acrylamide, N-amido(meth)acrylamide, N-acetamido (meth)acrylamide, N-tris(hydroxymethyl)methyl (meth)acrylamide, styrenesulfonic acid, homopolymers of styrenesulfonic acid, co-polymers of styrenesulfonic acid, N-(methyl)acryloyltris(hydroxymethyl)methylamide, (methyl) acryloylurea, vinyloxazolidone, vinylmethyloxazolidone, acrylic acid, methacrylic acid, vinyl sulfonic acid, styrene sulfonic acid, 4-acetoxystyrene (precursor of 4-hydroxystyrene), and vinylphosphonic acid, and vinyl methyl ether.

56. (Withdrawn) The composition of claim 55, wherein the synthetic polymer is poly(acrylic acid-co-N,N-dimethylacrylamide) or poly(N,N-dimethyl acrylamide-co-styrene sulfonic acid).

57. (Withdrawn) The composition of claim 53, wherein the synthetic polymer comprises a copolymer, wherein the copolymer comprises at least one monomer chosen from allyl amide hydrochloride, (3-acrylamidopropyl)trismethylammonium chloride, N-(3-aminopropyl)methacrylamide hydrochloride, and N-vinyl amides hydrolyzed to give an amino group.

58. (Withdrawn) The composition of claim 57, wherein the synthetic polymer is poly(N-(3-aminopropyl)methacrylamide-co-N,N-dimethylacrylamide).

59. (Withdrawn) A system for biological separation, the system comprising:

polyelectrolyte material wherein the polyelectrolyte material is adapted to coating ion-exchange material and to providing sieving for separation of at least one of PCR reaction products or DNA sequencing reaction products.

60. (Withdrawn) The system of claim 59, wherein the system further provides desalting.

61. (Withdrawn) The system of claim 59, wherein the system does not provide desalting.

62. (Withdrawn) The system of claim 59, wherein the system the ion-exchange material comprises cationic ion-exchange material and anionic ion-exchange material.

63. (Withdrawn) The system of claim 62, wherein the system is in the form of a mixed bed.

64. (Withdrawn) The system of claim 63, wherein the cationic ion-exchange material and the anionic ion-exchange material are present in stoichiometrically equivalent amounts.

65. (Withdrawn) A particle for biological separation, the particle comprising:

polyelectrolyte material wherein the polyelectrolyte material is adapted to coating ion-exchange material and to providing sieving for separation of at least one of PCR reaction products or DNA sequencing reaction products,

wherein the polyelectrolyte material comprises at least one polyanion chosen from poly(styrenephosphoric acid), poly(phosphoric acid), homo-polymers of maleic acid, co-polymers of maleic acid, homo-polymers of fumaric acid, co-polymers of fumaric acid, peptide polyanions, poly(aspartic acid), poly(galactronic acid), poly(glutamic acid), nucleic polyanions, poly(adenylic acid), poly(inosinic acid), poly(uridylic acid), and polysaccharides.

66. (Previously Presented) The method of claim 21, further comprising coupling the core with at least one PCR reaction product chosen from primers, primer-dimer, ssDNA fragments, unincorporated nucleotides, and salts.

67. (Previously Presented) The method of claim 21, wherein the particle is adapted to substantially exclude dsDNA fragments having greater than 100 basepairs.

68. (Currently Amended) The method of claim 21, wherein the core comprises [[of]] a porous ion-exchange material.

69. (Previously Presented) The method of claim 68, wherein the ion-exchange material is surface-activated.

70. (Previously Presented) The method of claim 68, wherein the ion-exchange material has a pore size of 100 Angstroms to 2000 Angstroms.

71. (Previously Presented) The method of claim 69, wherein the polyelectrolyte copolymer material has a Mw of 1.0 megaDaltons to 3.0 megaDaltons.

72. (Previously Presented) The method of claim 71, wherein the ion-exchange material has the pore size of 1000 Angstroms and the polyelectrolyte copolymer material has Mw of 1.7 megaDaltons to 2.4 megaDaltons.

73. (Previously Presented) The method of claim 21, wherein the polyelectrolyte copolymer material comprises polyanions and polycations.

74. (Previously Presented) The method of claim 73, wherein the polyanions and polycations form alternating layers.

75. (Cancelled)

76. (Previously Presented) The method of claim 45, further comprising coupling the core with at least one DNA sequencing reaction product chosen from primers, dye-labeled primers, nucleotides, dye-labeled nucleotides, dideoxynucleotides, dye-labeled dideoxynucleotides, and salts.

77. (Previously Presented) The method of claim 76, wherein the particle is adapted to substantially exclude dye-labeled ssDNA fragments having greater than 45 nucleotides.

78. (Currently Amended) The method of claim 45, wherein the core comprises [[of]] a porous ion-exchange material.

79. (Previously Presented) The method of claim 78, wherein the ion-exchange material is surface-activated.

80. (Previously Presented) The method of claim 78, wherein the ion-exchange material has a pore size of 5 Angstrom to 1000 Angstroms.

81. (Previously Presented) The method of claim 80, wherein the polyelectrolyte copolymer material has a Mw of 1000 Daltons to 6.0 megaDaltons.

82. (Previously Presented) The method of claim 81, wherein the ion-exchange material has the pore size of 10 Angstroms to 50 Angstroms and the polyelectrolyte copolymer material has Mw of 2.4 megaDaltons to 4.9 megaDaltons.

83. (Previously Presented) The method of claim 45, wherein the polyelectrolyte copolymer material comprises polyanions and polycations.

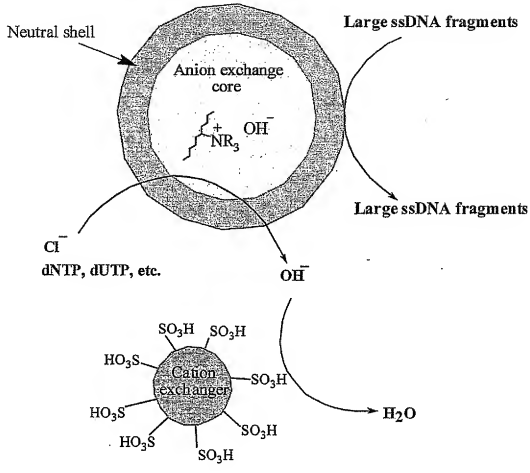
84. (Previously Presented) The method of claim 83, wherein the polyanions and polycations form alternating layers.

85. (Cancelled)

86. (Previously Presented) The method of Claim 21, wherein the polyelectrolyte copolymer material has a composition in molar percent for the at least one type of charged monomer of between about 0.1 percent to about 20 percent.

87. (Previously Presented) The method of Claim 45, wherein the polyelectrolyte copolymer material has a composition in molar percent for the at least one type of charged monomer of between about 0.1 percent to about 20 percent.

Fig. 1. Size-Excluded Anion-Exchange (SEAE) Based on a Core-Shell Structure According to Various Embodiments



- **Anion-exchange core to scavange small anions**
- **Crosslinked gel shell to exclude large fragments**

Copolymerization of Acrylic acid and N,N-Dimethylacrylamide

Formulation:

Ingredients:	FW	mL	wt (g)	m mol	wt %	monomers mmol	monomers molar %
Water (Milli-Q)	18.015	200.0	200.0	11101.860	95.9536%		
N,N-dimethylacrylamide (Dajac)*	99.130	--	8.0362	81.067	3.8555%	81.067	94.80%
Acrylic acid (99%, Aldrich Chemical)**	72.060	--	0.3202	4.444	0.1536%	4.444	5.20%
TEMED (d 0.7700, ultra pure, Amresco)	116.21	0.080	6.16E-02	5.30E-01	0.0296%		
APS (99.99 %, Aldrich Chemical) ***	228.196	0.800	1.60E-02	7.00E-02	0.0077%		
			208.43	11187.97	100.00%	85.51	100.00%

(Measured quantities in red color)
* High purity, 76 ppm MEHQ
** containing 200 ppm pf MEHQ
***Aqueous solution of APS (1.9971 wt%): 0.45158 g of APS in 22.1566 g of water (Milli-Q)

Feed Ratio:	(parts:100 parts)	(%)	
Monomers : water	4.18	4.01%	
[AA] : [DMA]	5.481	5.20%	(3:10 molar feed ratio; ~ 2:10 in copolymer by calculation)
[APS] : [Monomers]	0.082	0.082%	
[TEMED] : [APS]	757.105	88.333%	

Conditions:

Reaction vessel: 500-mL round bottom flaske with three 24/40 ground glass joints, equipped with a perforated 2" Teflon blade, a rubber septum witha glass bleeding tube for dry hellum purging on one joint, and a rubber septum with a 12 gauge syringe needle for venting on the other.
Stirring speed: 200 rpm
Reaction temperature: ambient temperature in a water bath
Deoxygenation: carried out in reaction vessel at ambient temperature with ultra pure helium (99.999 % pure) at 150 mL/m for 30 minutes and mechanical stirring at 200 rpm.
Inert atmosphere: reaction mixture bubbled with ultra pure helium (99.999 % pure) at 70 mL/m and constant mechanical stirring at 200 rpm.
Oil bath temperature: 45 ± 1 °C
Polymerization time: 15 hours
Addition of APS and TEMED: after deoxygenation, prior to lowering of the reaction flask into the oil bath.
Dialysis: 4 days in a 5-Gal PP tank in Spectra/Pro-7 membrane with 50K MWCO, water-changed once a day

Workup Procedure:

After 15 hours, the reaction mixture was a viscous solution. It was dialyzed and freeze dried to give 7.70 g (92.2 % yield) of polymer.

M_w = 3,395 kDa
M_n = 2,672 kDa

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Polyanion Ionic-interaction with Anion Exchanger

							Passive Polyanion Coating					
						Control	1	2	3	4	5	6
#	Anion Exchange Resin	Matrix type	Pore Size (Å)	Capacity (meq/mL)	pI, (wet)	Bare Resin	0.50 wt% Poly(DMA-co-AA) 3400K Da 3.8 wt% AA					Sheared salmon sperm DNA (cont AX & MB)
A												
B	Paroline-Chromalith	?	?	1400	20.0							
C												
D	Macro-Prep-Hi Q	PMMA	1000	400	15.0							
E												
F		PS/DVB	10-15	1350	15.0							
G		PS/DVB	10-15	1350	20.0							
H	Amicon A-27	PS/DVB	10-15	1400	15.0							

Key:

Good Results.

Procedure:

(1) To 0.20-0.25 mL of wet anion exchange resin in a 1.5 mL Eppendorf Microcentrifuge Flex-Tube (polypropylene), 1 mL of Milli-Q water was added, vortexed, spun, and supernatant removed. This series of steps was repeated 2 more times

(2) To the resin pellet, 1 mL of 2.0 M NH4OH was added, vortexed, 5 min standing at ambient temperature, spun, and supernatant removed. This series of steps was repeated 2 more times

(3) To the resin pellet, 1 mL of Milli-Q water was added, vortexed, spun, and supernatant removed. This series of steps was repeated one more time.

(4) To the resin pellet, 0.5 mL of Milli-Q water was added, vortexed to give a suspension, and stored in a refrigerator prior to use.

(5) To a 1.5 mL Microcentrifuge tube (polypropylene) containing 15-20 µL of the wet anion exchange resin in hydroxide form, 1 mL of the polymer solution was added.

(6) It was vortexed for 1 minute, let standing at ambient temperature for 5 minutes, vortexed for additional one minute, spun down in a centrifuge, and supernatant removed.

(7) The polymer solution coating was repeated one more time. The pellet was then washed with 1 mL of DI water and spun down. The DI water wash was repeated 2x.

(8) The final washed resin was re-dispersed in 500 µL of DI water and stored in a refrigerator prior to use.

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